PATENT

DOCKET NO.: TIBO-0008
Application No.: 09/640,787

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of:
Brendan Larder, Sharon Kemp, Stuart Bloor and Ann Brophy

Confirmation No.: 7344

Application No.: 09/640,787

Group Art Unit: 1648

Filing Date: August 18, 2000

Examiner: Ulrike Winkler

For: Method for Mutation Detection in HIV-1 Using POL Sequencing

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Sir:

DECLARATION UNDER 37 C.F.R. § 1.132

I, Paula McKenna, declare as follows:

1. I hold the position of Director Diagnostic Lab Operations Virco BVBA (Virco), Generaal de Wittelaan L11/3, B2800 Mechelen, Belgium.

2. Prior to my position as a director at Virco BVBA, I spent five years in clinical drug development with Johnson and Johnson Europe.

3. I received my Ph.D. in virology from Queen's University of Belfast, N. Ireland. A copy of my *curriculum vitae* is attached as Exhibit A.

4. I spent three and a half years as a postdoctoral fellow at The Belgian Zoonosis Workgroup studying epidemiology of Hantaviruses. I have over three years of experience in amplicon generation and sequencing.

5. I have read and am familiar with the contents of the above-reference patent application. I further understand that the nature of the rejection at issue in the pending application is that the examiner believes that the pending claims are obvious in view

of Herrogs *et al.* in view of any one of Zazzi *et al.*, Kozal *et al.*, Birk *et al.*, Cabana *et al.* Demeter *et al.* or Boden *et al.*

6. I further understand that the examiner requested in Paper No. 21, that if Applicants' specific sequences produce unexpected results, then Applicants need to point out what those unexpected results are in view of ¶6.

7. The purpose of this declaration is to address the issue described in ¶6 and ¶7 and as I will explain below, the primers and methods using these primers, at minimum, produce unexpected results and are therefore distinguishable over the primers and methods in the cited references in view of the unexpected results described herein.

8. The amplicon generated by the Virco primers is as follows. The PCR amplicon generated covers parts of two adjacent genes (polyproteins): GAG and POL over a total length of 1868 base pairs (bp). The POL region sequenced by Virco includes amino acids 1 through 99 of the protease gene and amino acids 1 through 400 of the reverse transcriptase gene.

9. The amplicon described in ¶8 above contains the last 347 bp of the GAG gene (out of a total of 1503bp) and the first 1768 bp of the POL gene (out of a total of 3012bp). The sum of these two stretches is larger than 1868 bp because there is a frame-shift and an overlap in both reading frames of 247 bp.

10. In our assay, we focus on identifying mutations appearing in the protease and the reverse transcriptase genes because protease and reverse transcriptase are the main targets of the antiretroviral drugs currently on the market: protease inhibitors (PIs),

nucleoside reverse transcriptase inhibitors (NRTIs) and nonnucleoside reverse transcriptase inhibitors (NNRTIs).

11. In comparing the primers with those of our competitors, the Virco primers enable us to detect important mutations associated with drug resistance, in particular mutations at positions 318 and 333 in RT which confer resistance to NNRTIs and NRTIs, respectively. See Harrigan *et al.*, 2002 ,. *Virol.* 76:6836-6840 (attached as Exhibit B) and Kemp *et al.* 1998, *J. Virol.* 72:5093-5098 (attached as Exhibit C). Surprisingly and unexpectedly, these mutations are not detected by others.

12. This means that the interpretation of the sequences of viruses tested using Virco primers produces a more accurate and complete resistance profile than other assays not covering that region.

13. Furthermore, the protease is one of the more important proteins for HIV as it cuts the polyproteins into separate functional entities (*e.g.*, the POL gene contains the protease, the reverse transcriptase and the integrase gene). As a result of antiretroviral therapy, mutations are selected that allow the virus to escape drug action. However, these mutations can cause structural alterations in the active site of the protease. It has been found that as a result, compensatory mutations at the protease cleavage sites themselves are in turn increasing in frequency during antiretroviral therapy, causing a further increase in drug resistance.

14. The Food and Drug Administration (FDA) has shown an increased interest in the effect of these primary protease mutations (in the functional cavities of the enzymes to overcome antiretroviral therapy (ART)) resulting in secondary

compensatory mutations (mutations in, *e.g.*, cleavage sites to overcome structural alterations in the active site of the protease).

15. The Virco assay and primers unexpectedly cover this FDA interest as the Virco amplicon includes a short partial GAG sequence containing two important cleavage sites for the HIV protease called "p7/p1" and "p1/p6". In this way we are able to monitor the evolution of the secondary mutations in the downstream part of GAG resulting from PI treatment.

16. Taken together, the foregoing primer characteristics establish that the Virco primers for amplicon generation and sequencing show unexpected results.

17. As evidenced by the unexpected results of the Virco primers, I believe that the Virco primers of the present invention would not be obvious to one skilled in the art, such as myself, when looking at the references as cited in Paper No. 21 and known competitors.

18. I further declare that all statements made herein of my knowledge are true and that all statements made on information or belief are believed to be true; and further that these statements are made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code, and that any such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Date: 24th May 2004

Paula Mc Kenna

Paula Mc Kenna, Ph.D.

Exhibit A: *Curriculum vitae* of Paula McKenna
Exhibit B: Harrigan *et al.*, 2002, *Virol.* 76:6836-6840
Exhibit C: Kemp *et al.*, 1998, *J. Virol.* 72:5093-5098